


***Chaetomium novozelandicum* PDD 97387  
(Holotype = AEB 1071)**


This page and the next two update the pdf I prepared in 2009 for *Chaetomium subaffine* AEB 1071. Pages that follow these provide pictures, descriptive detail and culturing results that were prepared in 2008/2009. Cultures of the + and – strains and their mated culture were sent to CBS (Centraalbureau voor Schimmelcultures) in 2009:

<u>CBS accession nr.</u>	<u>Your strain nr.</u>	<u>Name</u>
CBS 124555	AEB 1071 +	Chaetomium subaffine
CBS 124556	AEB 1071 -	Chaetomium subaffine
CBS 124557	AEB 1071 +/-	Chaetomium subaffine

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**28 February 2021 designations online:**

 CBS 124555 *Chaetomium novozelandicum* X.W. Wang, Crous & L. Lombard, *Persoonia* 36: 110 (2015) [MB#81]

 CBS 124556 *Chaetomium novozelandicum* X.W. Wang, Crous & L. Lombard, *Persoonia* 36: 110 (2015) [MB#81]

**CBS 124555**

**Taxonomy**

Taxon name: *Chaetomium novozelandicum* X.W. Wang, Crous & L. Lombard, *Persoonia* 36: 110 (2015) [MB#812980]

Name changes: *Chaetomium subaffine*

Status of the strain: ex holotype of *Chaetomium novozelandicum*

Type of organism: Filamentous fungi

**Collections**

Herbarium number: AEB 1071, holotype; CBS H-22191, isotype

**Origin**

Substrate (including host): Dead unidentified decaying twigs

Country: New Zealand

Locality: W. coast of the N. Island; town of Otaki; compost pile at 83 Atkinson Ave.

Collected, Isolated & Deposited by Daniel P. Mahoney

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**Reference within which the new species was published:**

Wang XW, Lombard L, Groenewald JZ, Lil J, Videira SIR, Samson RA, Liu XZ, Crous PW (2016) Phylogenetic reassessment of the *Chaetomium globosum* species complex. *Persoonia* 36:83–133. <https://doi.org/10.3767/003158516X689657>

**Continued on the next page**

**From page 110 of the reference given on the preceding page:**

***Chaetomium novozelandicum*** X. Wei Wang, Crous & L. Lombard, sp. nov. — MycoBank MB812980

**Etymology.** Refers to the country New Zealand, where this fungus was first collected.

**Cultures sterile.** *Chaetomium novozelandicum* forms a unique lineage (Group IIC, Fig. 1), basal to the *C. globosum* clade. This species differs by fixed unique SNPs in five loci: *rpb2* positions 3(C), 9(C), 12(C), 24(A), 39(C), 51(A), 60(T), 69(T), 99(C), 124(T), 138(A), 177(G), 186(C), 220(A), 300(C), 306(A), 312(A), 372(G), 376(T), 393(T), 420(C), 450(A), 525(T), 570(T), 573(C), 579(G), 582(G) and 597(T); *tub2* positions 12(C), 28(G), 97(T), 102(indel), 109(A), 142(indel), 143(indel), 144(indel), 168(C), 235(G), 236(G), 278(C), 319(T), 322(indel), 343(T), 368(A), 375(T), 378(A), 387(C), 447(indel), 459(C), 509(T), 570(G), 579(G), 656(T) and 707(T); *tef1* positions 262(A), 284(T), 396(C), 465(C), 519(T), 683(C), 744(C), 762(T), 816(C) and 870(C); *rpb1* positions 44(T), 59(C), 110(indel), 111(indel), 117(G), 163(C), 166(A), 175(G), 211(C), 256(C), 268(C), 272(A), 316(C), 364(T), 418(T), 427(G), 455(A), 457(C), 463(T), 487(C), 523(C), 535(T), 556(T), 580(G), 592(A), 613(C), 676(C), 685(T), 721(G) and 724(C); ITS positions 142(C) and 452(C).

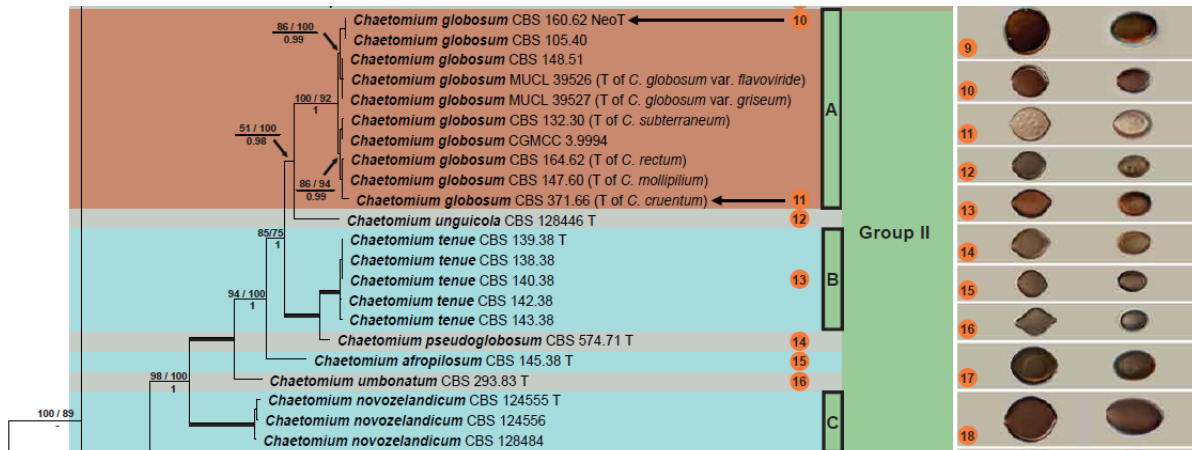
**Culture characteristics** — Colonies on OA with white, floccose aerial hyphae, without coloured exudates; reverse uncoloured.

**Materials examined.** NEW ZEALAND, town of Otaki on west coast, isolated from dead unidentified, decaying twig in a compost pile, collection date unknown, *D.P. Mahoney* (holotype AEB 1071, isotype CBS H-22191, culture ex-isotype CBS 124555); same collection details, CBS 124556. – USA, California, isolated from scalp of *Homo sapiens*, deposited in CBS by *D.A. Sutton*, 29 Sept. 2010, CBS 128484 = UTHSC 08-1518 = dH 21631.

**Notes** — Both phylogenetic inference and SNP analysis indicate that *C. novozelandicum* represents a novel phylogenetic species basal to Group II (Group IIC, Fig. 1). All attempts to induce sporulation on OA failed, even with the addition of sterile elm twig pieces.

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From a portion of Fig. 1 on page 88 of the reference describing the new species:



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Fortunately, CBS had safe-guarded viable + and – stains of this heterothallic species but, unfortunately, the new species was based solely on the sequences these cultures enabled. No teleomorph or anamorphic detail was presented because the strains sequenced had gone sterile. The following pages provide pictures, descriptive detail and culturing results that I prepared from fertile material in 2008/2009.

## ***Chaetomium subaffine* – AEB 1071**

This isolate from compost pile decaying twigs is clearly a member of the *C. globosum* Kunze complex. Members of the complex all share similar perithecial shapes, an intricata or intricata/epidermioid peridium, unbranched verrucose terminal hairs, 8-spored stipitate clavate asci and broadly limoniform, biapiculate, bilaterally flattened, brownish ascospores with numerous droplets and single apical germ pores. They vary in their maturation times, amounts of aerial mycelium in culture, exudate and terminal hair coloration, degree of terminal hair straightness, undulation, twisting or spiraling, perithecial and ascospore sizes, homothallism or heterothallism and the presence or absence of an anamorph (spermatial) state.

The most recent expansive treatments of the genus (Arx et al., 1986; Doveri 2008) separate *C. subaffine* and *C. angustispirale* Serg. from the *C. globosum* complex – based on slightly larger ascospores and perithecia but especially on their heterothallism and the presence of a conidial (spermatial) state. Of these the less well known *C. angustispirale* (known only from the type) is distinguished by its larger perithecia bearing more spiraling terminal hairs and by its slightly larger ascospores. According to Arx et al. (1986), Sedlar et al. (1973) and Müller & Sedlar (1977) discovered the heterothallism of *C. subaffine*. Others, however, including the original description by Sergegeva (1961), describe only the teleomorph and make no mention of an anamorph or any heterothallism. Field collections don't seem to be accompanied by any anamorph and cultures of these collections are often not attempted.

Despite the amount of work done on this important genus, difficult species complexes within the genus remain – such as the numerous species now lumped within or only marginally separated from *C. globosum*. CBS is now conducting extensive sequencing of its *Chaetomium* isolates. Hopefully this will help clarify relationships that morphological characters have failed to distinguish. I am in hopes that CBS will sequence my heterothallic *C. subaffine* to that end, especially as a help in clarifying its relationship to other isolates of *C. subaffine* and their relationships to the *C. globosum* complex as a whole.

**Important cultural and morphological characters that distinguish my isolate of *C. subaffine* include the following:**

1. Perithecial maturation in 17 days (25 C) on Difco CMA to which sterilized whole wild rabbit droppings were added (fruiting only on the droppings); no fruiting on CMA alone; fruiting by 21 days (and much later) on CMA to which several sterilized filter paper squares had been added (fruiting only on the filter paper); late fruiting also on potato carrot agar (our unfiltered preparation with many small potato and carrot fragments).
2. Abundant white floccose aerial mycelium on the CMA rabbit droppings and potato carrot agar cultures.
3. Growth rates similar to those recorded for *C. globosum* (Arx et al., 1986).
4. Any exudates clear with some yellowing of the filter paper and perithecial terminal hairs.
5. Heterothallic with conidia (spermatia) on all media employed as well as on all cultures inoculated with single or multiple ascospores. The conidia appear to be better described as annelloconidia or *Scopulariopsis*-like rather than phialoconidia.
6. Perithecial size and shape similar to those reported for *C. subaffine* (Sergegeva 1961, Arx et al. 1986). Perithecia slightly larger than those reported for *C. globosum* in Arx et al. (1986) and in most other references for that species but with terminal hairs flexuous, wavy and twisting similar to those illustrated for *C. globosum* in Arx et al., plate 33 and not as straight as those illustrated for *C. subaffine*, plate 77 (the latter is based on ATCC 22132 which is now recorded as *C. globosum* var. *rectum* on the CBS database - CBS 724.84). The original illustration by Sergegeva (1961) also shows straighter hairs than my isolate.
7. Peridium a textura intricata; asci and ascospores (except for slight differences in size) like those generally described for members of the *C. globosum* complex.

**Information on my collection of *C. subaffine* (AEB 1071) including its description in axenic culture:**

**Substrate:** small and medium-sized (<2 cm in diam) moist decorticated decomposing twigs of various garden plants. A majority of twigs, roughly 10 of 15 examined, exhibited this same species.

**Collection site:** deep in a 6–12 month old compost heap containing miscellaneous plant (garden) waste plus some chicken and horse dung; Otaki, N. Island of New Zealand

**Collector and identifier:** Dan Mahoney

**Collection date:** 13 November 2008

**Voucher materials:** Dried 72 day axenic Difco CMA + sterilized filter paper strips (mating of 3 plus strains – single ascospore cultures A, C & E and 2 minus strains – single ascospore cultures B & D), incubated at 25 C under alternating 12 hr light and 12 hr dark (dried culture designated AEB 1071); 2 Shear's mounting fluid (SMF) slide mounts; numerous digital photos of perithecium, perithecial contents and *Scopulariopsis*-like anamorph (spermatial stage); cultures [1. a culture originating from multiple-germinating ascospores. 2. cultures originating from single-germinating ascospores (single ascospore cultures A, C & E labeled plus strains; single ascospore cultures B & D labeled minus strains and cultures labeled plus strain (A, C & E together) and minus strain (B & D together) and 3. a mating culture that includes all 5 (A–E) single ascospore strains].

**See description in axenic culture on the next page**

**Description in axenic culture:** This description is based on axenic cultures originating from multi-ascospore germinations and to a lesser extent on the single ascospore plus × minus strain crosses on Difco CMA + sterilized filter paper squares. Incubation was at 25 C under alternating 12 hr. light – 12 hr. dark.

**Perithecia** especially numerous on potato carrot agar (PCA) ‘the multi-ascospore colonies especially’, on Difco CMA + sterilized rabbit droppings (on the droppings only) and on Difco CMA + sterilized squares of filter paper (on the paper only) - less aerial growth on the latter made the perithecia there more easily observed. Perithecia very similar to those described for *C. globosum* in Arx et al. (see their Plate 33, B) but matured more slowly and had more aerial mycelium. Perithecia on the rabbit droppings matured first and were overall ‘most natural’ - nearly mature after 16 days with mature ascospores by 17 days. Perithecia on PCA and on the filter paper of the Difco CMA culture weren’t mature until 3 weeks when numerous asci were still maturing. Perithecia (excluding the hairs) ellipsoid or broadly ovoid, 285–350 µm wide × ca. 400 µm high (height measurements are less meaningful due to the numerous dark hairs near the ostiole). **Hairs** mostly simple, septate, shorter and straighter on lateral portions of the perithecia, longer in terminal regions nearer the ostiole, roughened with numerous, small, evenly distributed ‘blisters’, pigmentation brown to grey brown in slide mounts with predominant terminal hairs grayish in reflected light or somewhat yellowish to faintly yellowish green as perithecia reach maturity and before ascospores, exuding in mass, obscured the hairs. The filter paper also becomes light yellowish as the perithecia mature and age. This yellowish coloration of filter paper strips also occurred in single strain cultures unassociated with perithecia. Any exudates are colorless to very faintly yellow. **Lateral hairs** roughly 2.5-3 µm wide at the widest point near their base. **Terminal hairs (nearer the ostiole)** straighter below but flexuous, wavy or undulate above (to a maximum of 4.5 µm near the base, narrowing to ca. 3 µm for much of their length, less pigmented near their apices and narrowly rounded apically). **Peridium** a brown *textura intricata* which can be clearly seen when, in squash mounts, the stiff hairs overlying it break away close to their bases – the circular dark hair scars are obvious among the *intricata* pattern of the outer peridium. **Asci** numerous, clavate with a reasonably long stalk, 8-spored, spore portion of the asci approximately 37.5–55 × 15–17.5 µm, deliquescing before ascospores are fully mature (or pigmented).

**Mature ascospores** broadly limoniform, smooth, moderately thick-walled, brown in transmitted light (dark in mass), biapiculate (usual view), bilaterally flattened (less frequently seen and this view more broadly ellipsoid with the biapiculate aspect less obvious), with a germ pore at one end (spore germination was observed from one end only in all spores) – the other end very similar and also slightly lighter than other wall areas, indicating a thinner wall but no germ pore. Filled with small globules in water mounts of lightly pigmented recently mature spores; globules larger and fewer - but still numerous – in fully pigmented spores; globules disappearing in older spores. When younger unpigmented spores (from whole sterilized rabbit droppings on an axenic Difco CMA culture) were first observed in a 70% EtOH mount irrigated with Melzer's reagent, a definite dextrinoid reaction (with reddish ascospores) was visible. During later mounts in just Melzer's (using rabbit pellets and other sources – PCA & filter paper), I sometimes viewed a slight redness in young ascospores but usually only an iodine coloration. Some degree of dextrinoid reaction seems to be present but I wasn't able to view it clearly in most mounts. Mature ascospores 11–13 × 9–10 × 8–9 μm.

**Anamorph (or spermatial stage)** *Scopulariopsis*-like, common among the aerial and surface-substrate hyphae, annellophores simple, separate or in small groups (but never crowded), basally septate (rarely with an additional septum part way toward the apex), smooth, hyaline, vaseiform with the widest point approx.  $\frac{1}{3}$  up or simply tapering gradually from this broader point toward the apex, nearly cylindrical apically, variable in size – 10–24 × 2.5–3 μm. Conidia in chains of up to approx. 15, occasionally irregularly clustering at the chain apex, obovoid to longer obovoid or nearly fusoid, hyaline, smooth with narrow truncate bases and faint detachment scars, conidia mostly 3–4(–5.5) × 1.5–2 μm.



## ***Chaetomium subaffine* AEB 1071 photo legends**

1. Ascoma, axenic 21 da CMA + filter paper, Melzer's mt.
2. Hairs closeup, original collection, Shear's mounting fluid.
3. Peridium, intricata, broken hair stubs, axenic 16 da CMA rabbit dung, water then Melzer's.
4. Asci & immature ascospores, axenic 24 da CMA + filter paper, Melzer's mt.
5. Asci & immature ascospores, axenic 24 da CMA + filter paper, Melzer's mt.
6. Spores with internal globules and perithecial hairs, 3 wk CMA + filter paper, + × – cross (single ascospore strains), water mt.
7. Spores without internal globules, 72 da CMA + rabbit dung, multiple ascospore inoculum, water mt.
8. Ascospore germination (pretreatment 5 min 3% hydrogen peroxide), 24 hr CMA, aniline blue lactic acid plus coverslip on agar.
9. Ascospore germination (pretreatment 5 min 3% hydrogen peroxide), 24 hr CMA, aniline blue lactic acid plus coverslip on agar.
10. Anamorph in situ on aerial hyphae (no coverslip), 8 da CMA.
11. Anamorph in situ on 8 da CMA agar surface, aniline blue lactic acid plus coverslip.
12. Anamorph in situ (in air bubble) on 8 da CMA agar surface, aniline blue lactic acid plus coverslip.
13. Anamorph in situ on 5 da CMA agar surface, aniline blue lactic acid plus coverslip.
14. Conidia in situ on 5 da CMA agar surface, aniline blue lactic acid plus coverslip.

